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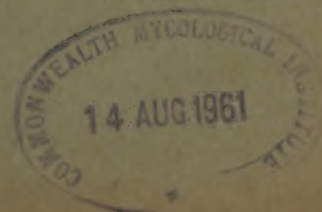
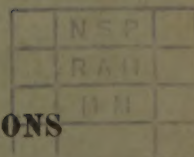
FAO Report No. 935

Report to the Government
of
YUGOSLAVIA
on
FRUIT TREE VIRUS DISEASES AND
THEIR CONTROL



FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS

ROME, 1958



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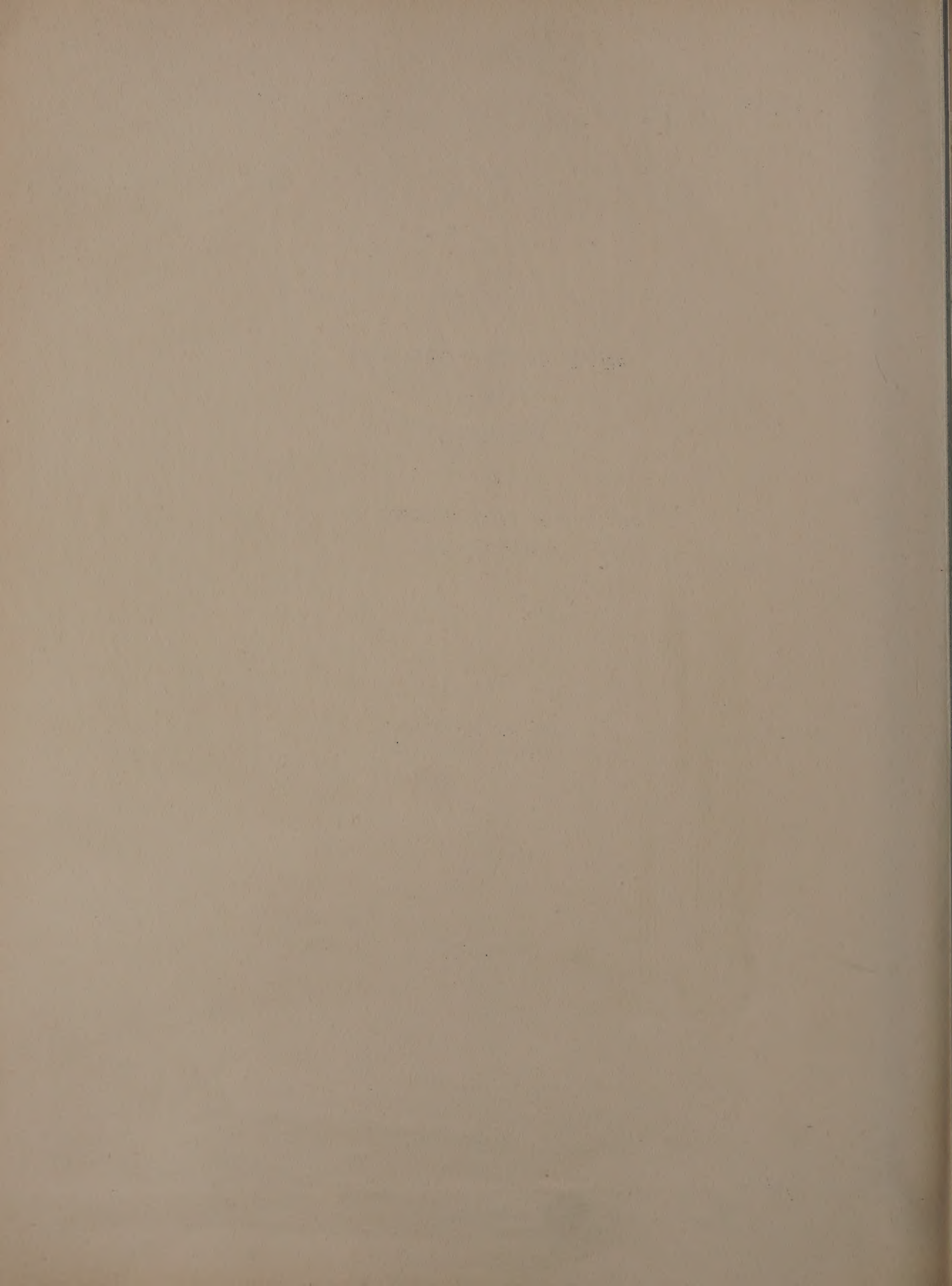
REPORT TO THE GOVERNMENT
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ON
FRUIT TREE VIRUS DISEASES AND
THEIR CONTROL

By

Lee M. Hutchins
FAO Plant Pathologist

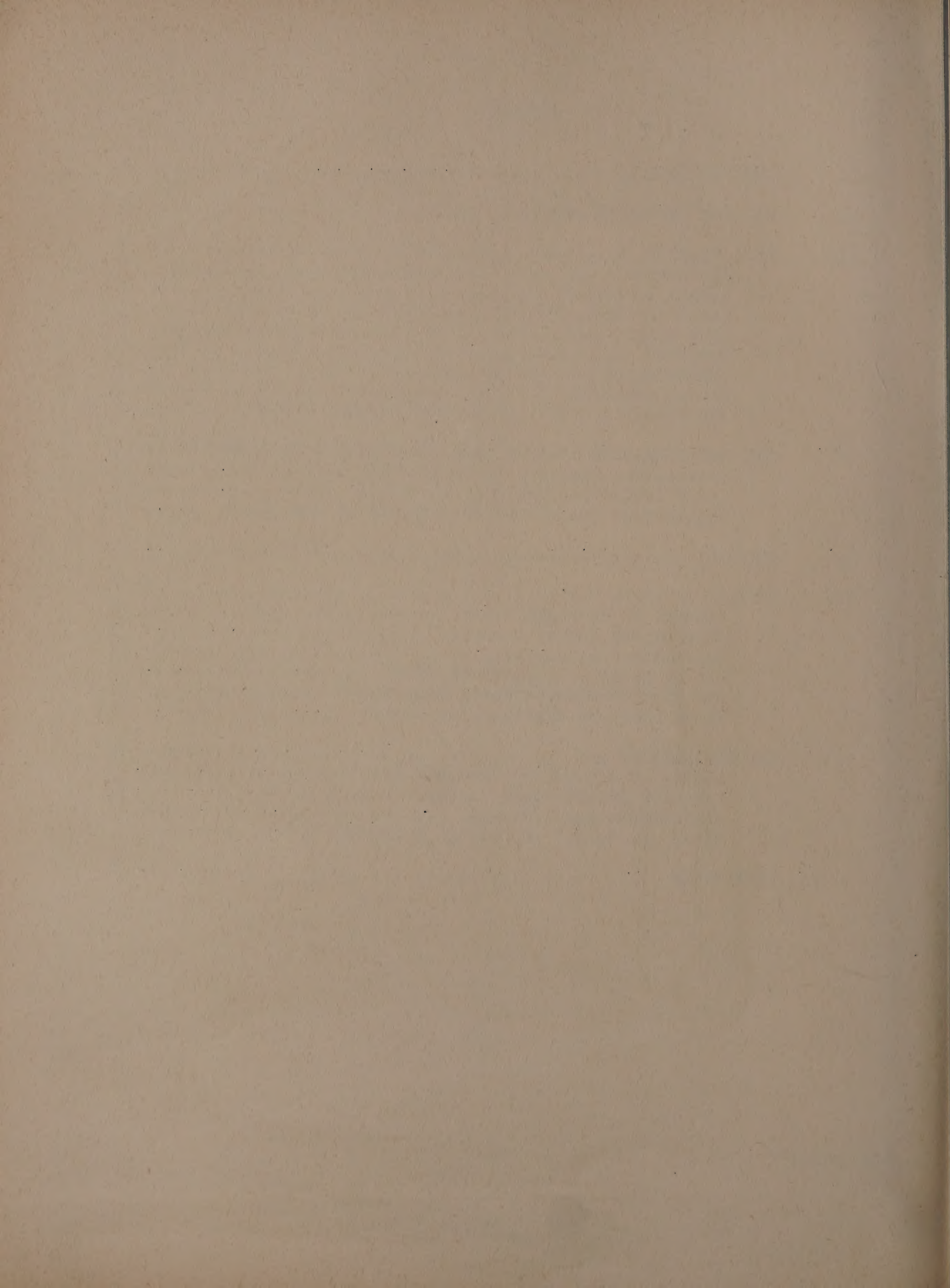
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I. INTRODUCTION

In accordance with the provisions of Supplemental Agreement No. 22 between the Government of Yugoslavia and the Food and Agriculture Organization of the United Nations, dated 11 March 1954, Dr. Lee M. Hutchins, an expert on virus diseases of fruit trees was appointed under the Expanded Technical Assistance Program to advise and assist the Government on the research and control of virus diseases of stone fruits, with particular emphasis on "sarka" disease of plums. The expert worked in Yugoslavia from 20 April to 15 December 1956.

The expert was stationed at the Institute of Plant Pathology, Faculty of Agriculture, University of Belgrade, Zemun. Visits were made to the leading fruit districts and experiment stations in the six Republics of Serbia, Croatia, Slovenia, Bosnia, Macedonia, and Montenegro. On each trip he was accompanied by one to three Yugoslavian plant pathologists or horticulturists, who were acquainted with the areas visited and the staffs at the research institutes and experiment stations, where conferences were held and orchard and nurseries inspected. On those occasions, the sarka disease and its behavior were thoroughly discussed, and other diseases of fruit trees also were considered. Accounts of these visits were included in monthly reports to the Government.

At the meetings of the National Extension Training Center, held at the University of Zagreb, 9-14 May 1956, the expert served as chairman and speaker at a workshop program entitled "Elaboration of an Extension Project in the Field of Plant Protection in Horticulture".

In October, the expert attended a meeting called by Mr. S. Todorović, in charge of the Federal Office of Plant Protection, to discuss and advise on plans for construction of a laboratory and glasshouse for the Faculty of Agriculture, University of Belgrade, for research on plant viruses and virus diseases. Photographs, descriptive notes, and architect's plans secured during the expert's visit to leading virus-research laboratories in England in July, and similar plans of new construction at Versailles, France, were exhibited and explained. Adaptations of these plans formed the basis for drawing up specifications to be submitted to architects of the University.

In November, the expert gave an evening program on Fruit Tree Virus Diseases in North America, before the Society of Plant Protection of Belgrade. About 30 virus diseases of grave concern in North America were discussed and illustrated with color slides. Emphasis

was placed on the diverse research methods required in proving widely different virus diseases of fruit trees; on control programs as they are organized and carried out in North America; on surveys, genetic resistance, and chemical identification tests; and on the possibilities as well as the limitations of methods for inactivating the virus in the host. A large part of the discussion was translated into Serbo-Croatian, and mimeographed copies were distributed to participants.

Attention was given to the training of Yugoslav students in connection with FAO fellowships. Information on facilities in universities and research institutions in Europe best suitable for study and training in plant virus diseases and research methods, together with a summary of similar opportunities for study in America, was made available to Government and University officials in Belgrade for use in preparing recommendations on FAO fellowship appointments.

Other activities included visits to the Institute of Plant Protection at Zagreb, the extensive experimental orchards of the State Orchard Farm near Varazdin, and the pesticide testing laboratory of the Zoeka Factory at Sabac.

At the conclusion of the assignment in November, two meetings were held in Belgrade for discussion of the work carried out and recommendations proposed by the expert during his service in Yugoslavia. The first was with Government administrative officials, under the chairmanship of Mr. Curuvija. Among the others present were Mr. S. Todorović, in charge of the Federal Office of Plant Protection; Prof. M. Josifović, head of the Institute of Plant Pathology at the Faculty of Agriculture, University of Belgrade; and Mr. John Howe, of the office of the Representative of the United Nations Technical Assistance Board. A résumé of the problems, of the work accomplished, and of the recommendations for future activities in research and control of sarka and other virus diseases of fruit trees in Yugoslavia was presented.

The second meeting, which was held at the Institute for Plant Protection at Belgrade, was with a technical group including research workers and program administrators. The principal discussion revolved around sarka disease of plum and how to control it.

The expert gratefully acknowledges the helpful, encouraging co-operation received from the Yugoslavian officials and research workers, which contributed immeasurably to the success of the undertaking. He is especially indebted to Ing. Mitar Jordović, specialist in sarka disease research at the Institute of Fruit Culture, at Čačak, for aid in carrying out the assignment.

II. THE PLUM AND PRUNE INDUSTRY

The plum and prune industry in Yugoslavia, next in importance to that in the United States of America, is seriously threatened by a virus disease called "sarka" or "plum pox". The word "sarka" means mottle and is descriptive of a characteristic color-pattern, symptom in the leaves. The term "plum pox" was used in an early description of the disease by Atanasoff (1) and refers to the shriveled, pocked appearance of the fruit.

To appraise the importance of sarka disease in Yugoslavia, one must have estimates on the extent of plum production and the value of plum products (2, 3, 4, 5).

Yugoslavia is reported to have more plum and prune trees than any other country in the world (4). In 1954, Yugoslav statistics (5) showed over 57 million plum and prune trees of bearing age, more than five times the number in California, and more than 20 times the combined numbers for Washington and Oregon. The distribution of bearing plum and prune trees in Yugoslavia, by Republics, in 1954 was as follows :

<u>Republics</u>	<u>Million trees</u>
Serbia	38.8
Bosnia	11.6
Croatia	5.0
Slovenia	0.8
Macedonia	0.7
Montenegro	0.5
total	<u>57.4</u>

The main plum growing areas are south of the Sava River and west of the Morava River in Serbia and in the northern part of the Drina River basin in Bosnia. Serbia, together with adjoining Bosnia-Herzegovina, the second most important region, accounts for about 88 percent of the country's total bearing plum and prune trees. The average annual yield for the years 1948-55 was in the neighborhood of 618,000 metric tons (Table 1).

Yugoslav statistics do not distinguish between prune and plum trees, but a distinction is sometimes made in descriptions of the fruit. All plum varieties that are freestones are treated as prunes, and all varieties that are clingstones are treated as plums. A

commonly accepted definition in the United States is that of Chandler (6) : "The prune of commerce is a plum that, because of its high sugar content and firm flesh, has been dried successfully without removal of the stone."

The plum and prune crop for the years 1948-55 is estimated by Jordović to have been distributed among various usages, as shown in Table 1.

Within Yugoslavia the average annual value of all plum products for 1948-55 was estimated at 88,545 million dinars (Table 1), or an equivalent of \$221,352,500. The total value of all exported plum products in 1955 was 2,034 million dinars, or \$5,085,000. By far the largest export item was dried prunes, valued at 1,828 million dinars, or \$4,570,000.

A Federal statistical report (5) shows that plum production in Yugoslavia far surpasses that of any other tree fruit. As compared with apples and pears, the next largest fruit crops, average annual production of plums during 1948-55 was 618,375 metric tons, or 72 percent of the total for those three fruits. Apples were second, with 160,000 metric tons or 20 percent, and pears third, with 70,000 metric tons, or 8 percent.

Table 1. Average value of plum products for 1948-55

Product	Production (metric tons)	Percent of total	Value (million dinars) ^a	Value of product exported in 1955 (million dinars)
Fresh fruit	32,395	5.2	648	158.0
Fruit made into brandy	479,680	77.6	71,952	15.4
Fruit made into jam	29,000	4.7	4,350	32.6
Dried fruit	77,300	12.5	11,595	1,828.0
Total	618,375	100.0	88,545	2,034.0

^a The average value in dinars of 1 kg (2.2 lb.) of each product is used in computing total values. Thus: 20 dinars per kg. of fresh fruit, 150 dinars per kg. of brandy, 150 dinars per kg. of jam, and 150 dinars per kg. of dried fruit. For comparison with dollars values of the products, the 1956 exchange rate of 400 dinars per dollar is employed; this is the exchange rate offered by the National Bank of Yugoslavia.

III. SARKA DISEASE

Importance

Sarka disease, or plum pox, is of major importance to plum growers in some of the most heavily planted districts of Yugoslavia. It has been found in all of the Republics but is most prevalent in Serbia, Macedonia, and some parts of Bosnia. It is estimated that 15 million of the country's total of 57.4 million bearing plum and prune trees, or 26 percent, are affected by the disease. By far the greatest number is found in Serbia, where 14 million trees, or 36 percent of the Republic's estimated total of 38.8 million are believed to be affected by sarka disease. The disease is also widely distributed in Bulgaria. Possibly it may occur in some of the countries bordering Yugoslavia and Bulgaria, but no reliable reports have been received.

Nature of losses

Sarka does not cause early death of the affected trees, but it has a deleterious effect on the fruits. Diseased trees produce a very inferior crop and orchards are ruined at an age when they should be most productive. Quantity as well as quality of fruit is affected. The fruits from diseased trees are smaller and weigh less than the fruits from normal trees. During the 1954-56 harvests, the affected fruits were lighter by 20 to 30 percent than the healthy fruits. Affected fruits contain about 1 to 3 percent less sugar than normal fruits. This reduces the quality of dried fruits, brandy, and jam. Brandy from affected fruits has a bad flavor and lower alcohol content. Drying of affected fruits is not profitable. About 30 percent of the fruits fall prematurely. Usually about 13 percent of the fallen fruits cannot be used at all. Tests show that normal-appearing fruits from diseased trees contain less sugar and are of inferior quality, in comparison with fruits from entirely healthy trees.

Cause of sarka disease

Transmission of sarka is easily effected by budding or grafting from diseased to healthy trees. The disease is not transmitted by injections of juice from infected parts of the trees, nor is it transmitted by seeds or through the soil. The incubation period following graft-inoculation is 9 to 13 months. Young trees show symptoms throughout the crown within 1 to 2 years after inoculation, but older trees may take several years to express symptoms throughout the crown.

Failure to find a visible organism associated with sarka, coupled with proof of graft-transmissibility and a known incubation period, places sarka in the group of graft-transmissible virus diseases.

Host range

Sarka is found naturally in plum and all cultivated plum varieties are affected, some more than others. It has been shown by artificial graft-inoculation that apricot, an ornamental plum (Prunus triloba), and myrobalan plum (P. cerasifera) also are susceptible. Myrobalan is used extensively in Yugoslavia as a root-stock for Požegača and other standard plum and prune varieties.

Christoff (7), working in Bulgaria, tried unsuccessfully to transmit sarka by graft-inoculation to almond, sour cherry, sweet cherry, Mahaleb cherry, peach and other genera. He budded from the test plants back to the susceptible Požegača plum, but did not obtain transmission. Likewise, Jordović in research soon to be published performed similar tests on sour cherry, Mahaleb cherry, peach, almond, pear and quince, but secured no transmission of sarka virus. It would appear that the virus has a limited host range and may be largely confined to plum; if true this would greatly simplify control undertakings. Of course, negative results of this kind need to be confirmed repeatedly before they can be accepted without qualification, and the tests are continuing.

Important additional inoculation tests are needed to determine whether wild Prunus species growing near plum orchards could harbor the virus and serve as reservoirs of inoculum with or without symptom manifestation.

Symptoms

In general, symptoms of sarka as observed in Yugoslavia are similar on all hosts. Different varieties of the susceptible species, however, exhibit some variation in symptoms, particularly in relation to the number, size, shape, and color of spots on the leaves.

In plum and prune, the disease is manifested in the leaves as pale-green or yellowish-green spots, rings, and lines. The spots or areas of chlorotic tissue are variable in size, and the margins usually are not clearly defined. Ring spots are very common and consist of a ring of chlorotic leaf tissue enclosing an area of green tissue. They are mostly 4 to 6 mm in diameter, and there may be 1 to 20 spots per leaf; rings are much larger in young than in old leaves. Lines of chlorotic tissue 10 to 15 mm. long and not in the form of rings are found along the main veins and sometimes along the secondary veins. On the fruit the disease causes lines or sunken areas which are rusty brown or pure brown. The flesh under these areas is mostly necrotic and gummy. Fruits of affected/tolerant varieties show darkening of the skin without lines or sunken areas.

Some plum varieties show severe symptoms on both the leaves and the fruit; examples are Požegača, Italian Prune, and King Dušan plum. Other varieties may show characteristic symptoms on the leaves but few to no symptoms on the fruit; examples are Moravka plum and an early red plum called Crvena Ranka. In myrobalan plum leaf symptoms are the same as those in the cultivated plum. Fruit symptoms, however, are different; a small number of the fruits have sunken lines. Usually this species is more tolerant to sarka than cultivated plum.

Transmission and spread

The problem of how sarka disease is disseminated has been controversial for many years. It is common knowledge that Atanasoff and Christoff, working in Bulgaria, considered the aphid Anuraphis padi to be an important vector of sarka virus. In 1956, Prof. I. Pobegajlo of the Faculty of Agriculture at Sarajevo stated that he held the same view and believed that one or more insect vectors are responsible for the rapid spread of sarka in Bosnia. He estimated, for example, that sarka spread from Sarajevo to Tuzla, a distance of 100 kilometers, in 7 years, or at the rate of about 15 kilometers per year. Vaclav, also of the Faculty of Agriculture at Sarajevo, stated that in limited tests in 1955 he was able to transmit sarka virus by another aphid, Phorodon humili.

Workers in Serbia, on the other hand, attach much less significance to insect vectors of sarka virus and believe that distribution of the disease is largely, if not entirely, brought about through unwitting use of infected suckers and scions in the horticultural propagation of plum trees in nurseries. Josifović (8), in charge of the Institute of Plant Pathology, Faculty of Agriculture, University of Belgrade, summed up the viewpoint of the Serbian pathologists in this way :

"According to Atanasoff and Christoff, 'plum pox virus' is transmissible by Anuraphis padi. Our observations in the course of 15 years allow us to conclude that this insect, possibly a vector of 'plum pox virus' under experimental conditions, can be active only to a small degree in nature. In a large number of orchards where from a few to many diseased prune trees are present, the disease has not progressed during the course of the last 10 to 15 years. In some prune hedges, aged 2 to 30 years, where trees are growing side-by-side, some trees are seriously attacked and others are free of the virus. In the same orchard, prune trees showing 'plum pox' are growing beside trees free of the disease." (Translated from the French.)

Continuing research on sarka disease and its means of natural spread possibly may harmonize these divergent interpretations. It is conceivable that although the disease appears to be spread mainly by horticultural propagation of the plum in Serbia, such means of spread in Bosnia and Bulgaria may be augmented by an effective insect vector that is of minor importance in Serbia.

Presence in fruit orchards of a damaging virus disease for which there is no evidence of a natural vector, or for which the evidence indicates that dissemination is principally by infected propagating materials, is somewhat unusual but not unknown. Familiar examples in North America are avocado sun blotch, black cherry canker, citrus exocortis, citrus psorosis, citrus stubborn disease, French prune diamond-canker, grape leafroll, and peach wart.

Some fruit tree virus diseases may show spread by a natural vector in one area and not in others. For example, little cherry has spread rapidly all through the Kootenay Lake area of British Columbia, where until now there are no uninfected sweet cherry trees. Reeves et al (9) reported that the virus has been found to be widely present in oriental flowering cherries (Prunus serrulata) in British Columbia and in many parts of the United States. Some of those infected but symptomless flowering cherries outside British Columbia, in the State of Washington for example, are in areas where sweet cherries are grown. But there is no evidence of spread in these last areas, even where the infected cherries have been maintained for several years adjacent to uninfected sweet cherry trees. The causal virus, however, is easily transmitted by grafting. It is concluded that rapid spread of little cherry in British Columbia is accomplished by a natural vector that is absent or is not effective in the adjacent State of Washington and other places in the United States where infected oriental cherries occurred.

Vector research

In vector research, care should be taken to use sufficient numbers of test plants so that the results of the transmission tests will be conclusive. Field plot techniques that permit statistical evaluation of results are a necessary part of all modern experimentation. It may be necessary to use as many as 100 plants in each determinative transmission test and to repeat such tests several times. With appropriate statistical contents the resultant data then can be interpreted directly in percentage values and reliable estimates of prevalence and effects on yields determined. If the insect is an efficient vector under conditions of the test, the data will be very convincing. If the insect is a weak vector, perhaps only 2 to 5 percent of the test plants will develop the disease. If too few plants are used in the tests, such

a small percentage of positive transmissions could be missed entirely; or, the efficiency of the insect as a vector could be greatly overrated because by chance several of the test plants happened to become infected.

Experience with vector transmissions of peach mosaic in the United States illustrates some of the difficulties of such research and the necessity for adequate determinative tests. It was reported from Colorado that peach mosaic in that State could be transmitted by the green peach aphid (Myzus persicae). Attempts to transmit the disease by this aphid in Texas and California, however, were unsuccessful. The solution of the problem came finally, in 1955; it was shown that in California the disease is transmitted by an extremely small Eriophyid mite (11), which feeds and develops under the bud scales. In these tests, if leafy, mosaic-diseased twigs on which aphids were feeding had been put in contact with the leaves and twigs of healthy plants and if the aphids had been allowed to crawl onto these plants and feed on them, transmission of the virus to the healthy plants could have appeared to take place as a result of the aphid feeding. Actually, it would have been accomplished by concomitant movement and feeding of the tiny Eriophyid mites, which are invisible to the naked eye.

Vector research is often best performed in insect-proof glass-houses or screened enclosures offering protection against accidental entrance of insects from outside. Special chambers should be provided for culture of virus-free plants and insects, for maintaining cultures of plants infected with the virus under study, and for exposing healthy plants to insects that have fed on infected plants. In some of the tests, approximate control of air temperature, humidity, and light may be important in order to provide favorable environment for transmission of the virus by the vector and for expression of disease symptoms in the successfully inoculated test plants.

Experimental plantings to test natural spread

Plantings to test the natural spread of sarka in heavily infected areas of Serbia have been made by Jordović. For the tests, healthy seedlings of the susceptible Požegaca plum were planted in orchard formation close to old orchards in which many of the trees were typically diseased. The size of the test plantings varies from 300 to 800 trees. Several locations were selected for the test orchards. Two of them are near Novi Pazar, 1 is at Prokuplje, and 1 is at Negotin. There had been no evidence of natural spread to the young test plantings by 1956, when they were about 2 years old.

These test plantings ultimately should provide conclusive evidence as to whether natural spread of sarka is taking place in the respective areas. Vector research is essential for determining species of insects that might transfer the virus from diseased to healthy plants, the conditions under which such insects are effective as vectors, and the host relationships. But such research performed under controlled conditions usually needs to be supplemented by adequate observations and tests in the field, in order to determine just what is taking place in nature. Answers to these questions have a very important bearing on control of sarka. Where the disease is spread mainly by horticultural propagation of plum, control will be principally through use of healthy propagating material. Where spread is taking place also through an insect vector, or such is suspected, control must include sanitation measures, such as removal of diseased trees, and also vector control where this is practicable. Whether vectors are present or absent, the test plantings such as Jordović has made in Serbia are needed to determine the spread behavior of sarka under field conditions. The results of these tests will have a direct bearing on the organization of a control program.

IV. NEED FOR PLUM-TREE REGISTRY AND BUD-CERTIFICATION SERVICES

It has been well demonstrated that measures against spread of sarka disease must include ways and means of insuring propagation of plum stocks in the nurseries from only healthy, virus-free parent trees. Helpful suggestions for accomplishing this may be found in the way a similar problem was worked out for controlling psorosis, or scaly bark, of citrus trees in California and in the stone-fruit-tree registration program in Oregon.

Citrus Tree Registry in California

Psorosis disease of citrus, the virus nature of which was determined by Fawcett (12), is widely distributed over the world. Trees harboring the virus may appear healthy for many years. Then, after a long incubation period, gross symptoms begin to appear in the form of large bark cankers, general dieback, and weakening of the trees.

There appears to be no insect vector of psorosis virus in California, and the disease does not spread naturally from tree to tree in nurseries or orchards. However, if buds or scions for nursery propagation of citrus are taken from trees apparently healthy but actually carrying the psorosis virus, the trees so propagated may grow normally for 10 or more years, after which incubation period they will develop scaly bark and go into decline.

For protection of nurseries and the citrus industry against this dread disease, the Department of Agriculture of the State of California, through its State Nursery Service, offers a valuable bud-certification service. This service was begun in 1937 at the recommendation of Dr. Fawcett and is known as the Citrus Tree Registry (13).

If a citrus tree harbors the psorosis virus, symptoms will usually develop before the tree is 15 years old. The California State Nursery Service conducts surveys for psorosis and registers productive, healthy trees that are 15 or more years old. Buds and scions from such trees will be free of the virus, and trees propagated from them will not develop the disease.

The State Nursery Service also inspects for psorosis citrus trees selected by nurserymen as sources of propagating material and reports results to each applicant. Registration of individual trees

is valid for 3 years (subject to immediate cancellation in the event of infection or an imminent threat of psorosis), and is renewable for another 3-year period.

The biological basis for registration has two essentials:

- (1) That the selected trees shall be more than 15 years old or provably derived from older trees available for inspection, and
- (2) that registry shall be based on absolute freedom from characteristic symptoms on bark and leaves at 15 points on each tree when examined at an appropriate season.

The losses from psorosis and the protection offered by the Citrus Tree Registry are described in a paper by Wallace (14).

Stone-fruit-tree Registry in Oregon

Tree registration, indexing for virus-free stone fruit trees, and the establishment of virus-free scion-wood orchards as currently practiced in Oregon have been described by Milbrath (15). In North America there are about 50 distinct virus diseases of stone fruits and nearly half of these occur in Oregon orchards. Tree Registration has greatly improved the quality of trees grown in nurseries and orchards. Virus-free scion-wood orchards and the use of virus-free propagating materials have several advantages. Varieties are more easily segregated and maintained true-to-name. A local source of fresh, virus-free bud-wood is always available, and no time is lost in collecting miscellaneous lots of bud-wood. Carefully maintained, virus-free scion-orchards will give a high percentage of vegetative buds and few flower buds. In the nursery, virus-free trees give a more uniform stand, a higher percentage of top-grade trees, and a lower percentage of degraded trees.

Suggestions for plum-tree registry in Yugoslavia

It would be highly desirable to organize a Plum Tree Registry Service in Yugoslavia along lines similar to the Citrus Tree Registry in California and the Stone-Fruit Tree Registry in Oregon. This service would provide an authentic source of virus-free propagating material such as budwood, scions, and suckers from parent plum and prune trees selected for productiveness, quality of product, and regional adaptability. Beginnings already have been made in this direction by Jordović in Serbia and Pobegajlo in Bosnia. The values that would accrue from the authenticity of varieties would go far to recompense any costs involved.

Great care should be taken in surveying the areas and in selecting the parent trees that are to provide virus-free propagating material. Inspections must be thorough and should be made when symptoms are at their best. Usually the most definite symptoms of sarka are seen in the spring and early-summer foliage and in the fruit during the ripening period.

Areas entirely free of sarka are the preferred locations for registering virus-free parent trees. Although the disease is widely distributed, such locations can be found. For example, in the course of a field inspection trip in the vicinity of Valjevo, Serbia, a visit was made to an isolated orchard consisting of 670 Požegaća trees 17 years old and in good condition. The orchard was supervised by the Agricultural High School at Valjevo. About half of the trees were in full fruit and in ideal condition for symptom expression. A tree-by-tree inspection was made by the expert and Jordović on 13 September 1956 and revealed no symptoms whatsoever of sarka or other virus diseases in the orchard. It is planned to reinspect the orchard 2 or 3 times during the growing season of 1957 as a further check on its healthy condition. If it comes through with a clean record, this orchard could constitute an exceptionally valuable source of clean material for nursery propagation of Požegaća trees.

Every precaution should be taken against accidental introduction of a virus disease into an orchard found to be virus-free and selected for tree registry. No stone-fruit trees should be planted adjacent to the orchard, and no replanting should be done except with proved virus-free nursery trees.

Trees selected for registration should be plainly and securely numbered and labeled. Their exact locations should be indicated on maps, and they should be frequently reinspected. The record should show the date of inspection, the name of the inspector, the variety, the age of the tree, its vigor and form, and its desirability from the standpoint of crop production. Some simple system of designation such as a ring of paint around the trunk of registered trees will facilitate locating them later, and is especially helpful where plantings are not in straight rows. A procedure found useful is to apply a belt of orange paint about the trunk, and after it has dried, paint the registration number in black on the band.

Progeny of registered trees should be closely observed in the nursery to be sure they are virus-free. Indexing should be freely used for the selected trees of plum varieties in which symptoms of sarka are mild or obscure. The indexing is done by grafting scions or buds from healthy Požegaća or other highly susceptible plum on the trees being considered for registry, or by grafting scions or buds from them on healthy trees of varieties that develop marked symptoms of sarka after being infected by the causal virus. If results of the indexing are negative, the trees so tested may be given registration numbers.

V. SURVEYS

Well-planned and thoroughly conducted surveys are highly necessary in appraising plant disease losses and are especially important for fruit-tree virus diseases.

Ordinarily the surveys are of two types: local and regional. Local surveys are adapted to detailed study and observation of specific diseases and are usually conducted or directed by research groups. Regional surveys over large areas provide information on distribution of the diseases, incidence by localities, rate of increase in different years, losses, and overall effectiveness of control measures where these are applied. Regional surveys are best performed by thoroughly trained inspectors under able direction, who work closely with the research groups.

In both regional and local surveys for fruit-tree virus diseases, information on seasonal behavior and symptom manifestation of a disease under different environmental conditions may be recorded. Also, susceptibility of different hosts and varieties may be observed, rate of spread determined, and economic losses evaluated. The information made available can provide the basis for control programs, including diseased-tree removal and quarantine measures where it is indicated that such action is warranted.

Annual surveys are systematically carried out in many of the large areas of stone fruit production in North America where virus diseases are a serious problem. In critical situations, the same areas are resurveyed one or more times each year. Accurate records are exceedingly important. The need for such surveys in the case of sarka disease in Yugoslavia has been pointed out by Josifović (16), Pobegajlo (17) and Todorović (18).

Aids to identification of virus diseases may be needed in survey work and in the selection of virus-free propagating material. This is especially true where visual symptoms are indefinite during unfavorable seasonal conditions, and in the case of tolerant varieties and species that seldom develop good symptoms. Indexing for sarka on susceptible varieties of plum, such as Požegača, has been mentioned. Recent techniques employing tissue inoculations on succulent plants such as cucumber and tomato under glasshouse conditions should be tried. If the trials are successful, positive symptoms of sarka might appear in the test plants within several days after inoculation. Chemical

tests for identification of sarka also should be explored. An example of the use of such technique is the acid-alcohol test for phony peach disease (19) in the United States, which proved to be specific for that disease. The test is rapid and easy to perform and is a valuable confirmatory aid to identification; it has been used extensively in the surveys for phony disease. . Discovery of a test of this character would be of advantage for sarka.

VI. CONTROL OF SARKA

Sarka virus is known to invade both the tops and the roots of infected trees. The most effective means of control, therefore, would be to destroy the diseased trees. If roguing is thorough, the source of inoculum would be greatly diminished and the hazard of spread by horticultural propagation of plums as well as by a natural vector of sarka virus would be correspondingly reduced. Another reason for removing the diseased trees is that the disease lowers both the productivity of the tree and the quality of the fruit. In most cases, removal of diseased trees would benefit the grower by enabling him to replant with healthy plum stock or use the land for a crop more profitable than diseased plums.

Removal of diseased trees

Plum trees in Yugoslavia are widely dispersed over a great number of small properties. Therefore, a general diseased-tree removal program for sarka disease, however commendable, would meet with many difficulties and delays. The program would be immediately important in areas where it may be shown that the disease is spreading from affected to healthy trees. Popularization of the program and its acceptance by the grower could be furthered by education through press notices and by visits from plant protection officials. The organization of local committees of growers would be helpful. Field meetings for demonstrating the disease and its infectious nature would be particularly convincing to the layman. Some of these demonstrations could well be staged on experimental plots for study of sarka at the research institutes and experiment stations.

Such means of bringing the problem to the public have been of great aid in programs for suppressing similar diseases in America. For example, in one instance it was considered necessary to remove phony-diseased peach trees on 1,200 properties in the State of Georgia, U.S.A. A preliminary campaign explaining the objectives and the methods was well carried out; subsequently, only 2 of the 1,200 property owners were unwilling to have their diseased trees removed. A State law authorizing removal of diseased trees by plant protection officers without the owner's consent was invoked only as a last resort in cases of this kind. It was done only where the occurrence of the disease was of unusual importance, and after persuasion had failed. Usually, upon visiting the demonstration plots at the research station, even those growers most opposed to diseased-tree removal were convinced of its merits.

The organization of surveys has been discussed. Often it is practicable for the survey crews to direct the removal of diseased trees. Occasionally an orchard may be so heavily diseased that the best procedure is to remove all the trees and replant with healthy plum stock or devote the land to other use. If it is decided to remove only trees showing the disease, the grower must be made to understand that under conditions of natural spread of sarka, only those trees showing identifiable symptoms can be removed at one operation. Other apparently healthy trees may be actually infected and have the disease in its incubation stage. These will not show symptoms until later and ordinarily must be removed at the next or later inspections.

If symptomless infections are not explained to the property owners, they may lose faith in the control program, or get the impression that the program somehow is increasing the number of diseased trees. They should be told that each removal operation reduced the reservoir of virus in the orchard and decreases the opportunity for spread. Repeated inspections and removal of diseased trees as symptoms develop may be expected to reduce markedly the incidence of new cases in the control area.

The capacities of a virus disease that has a very active insect vector to flare into serious proportions, and the effect of latent infections on prevalence of diseased trees, are well illustrated by peach mosaic (10) in an important orchard district in western Colorado. Seven mosaic-diseased trees were found in 1931. The number had increased to 6,507 diseased trees by 1934. Removal of trees in which mosaic could be identified was begun in the fall of that year. In 1935, however, there were 30,457 new cases of peach mosaic in these orchards, most of which were in the incubation stage the previous year and not showing symptoms at that time. Removal of most of the diseased trees in the spring of 1935 deflected the incidence curve downward. In 1936, there were 9,561 new cases, and in 1937 about 4,500 new cases. The number of new cases was further reduced, later.

Diseased-tree removal for control of phony-peach disease (19) was begun in 1929. In the course of extensive annual surveys, 35 million peach orchard trees had been inspected by 1932, and the disease was found to occur in southeastern United States from Virginia to Texas. From 1929 to 1950, about 1,600,000 phony-diseased trees were removed. The program of orchard inspection and diseased-tree removal soon resulted in a marked decrease in the total number of phony trees. In seven states where infection was light, the disease was apparently eradicated. In seven other states where infection was heavy, the incidence of diseased trees was drastically reduced.

In the United States, the most devastating outbreaks of peach yellows, peach rosette, little peach, red suture, phony disease, and peach mosaic have yielded to immediate removal and destruction of all visibly diseased trees when performed concurrently with frequent, efficient, well-timed inspections of orchards, home plantings, nurseries, and trees escaped from cultivation.

Total eradication of a virus from an area may be difficult or even impossible. In North America, control agencies prefer to say "suppression" of a disease. Only by continued vigilance year after year, in which surveys are followed by removal of diseased trees, can the "suppression" of sarka be expected to be effective in regions such as Bosnia, where the disease is regarded as spreading rapidly.

Controlling insect vectors

Earlier in this report, it was mentioned that certain species of aphids are reported to spread sarka virus from diseased to healthy plum trees in Bosnia. Pathologists and entomologists should be encouraged to determine the conditions under which these aphids are effective vectors in that region. In addition, the cultivated and wild plants on which the aphids may feed and the possibility that such plants may become infected and harbor the virus should be investigated. A thorough study is needed to determine the practicality of reducing spread of sarka through vector control and if vector control is practical the best procedures to follow.

Quarantine

Plant quarantine measures are commonly directed against movement of a new and dangerous plant disease into or within a country or a region. For several reasons it would appear impractical at present to enforce general quarantine regulations against sarka in Yugoslavia. The disease already is widely distributed in the country. More research is needed on the disease and its manner of spread in the different Republics, and as yet there is no organized program of control. However, full precautionary measures should be taken against further distribution and planting of nursery trees infected with sarka virus.

Use of healthy propagating stock

The need for bud-certification service, for plum-tree registry, and for establishment of healthy scion orchards was discussed under those topics. Such services would aid greatly in providing approved sources of virus-free budwood, scions, and suckers for propagating healthy nursery trees or for top-working orchard trees. Federal and local plant protection services no doubt could assist materially in furthering a program of this kind.

Use of disease-resistant plum trees

The Agricultural Research Institutes at Sarajevo and Cacak are engaged in research to determine the tolerance of many varieties and selections of plums, prunes, and related stone fruits to the disease. The experimental trees are planted in orchard formation so that their value as crop trees also can be observed.

After the trees have become established and are growing well in the experimental orchard, some trees of each variety are inoculated with sarka virus by grafting on them a scion from a diseased Požegača tree. Sarka virus from the infected scion moves through the graft union and invades the branches of the test tree, where the virus multiplies, becomes distributed, and induces symptoms of sarka that are characteristic for infected trees of the variety under test. These symptom manifestations then are easily compared with those of the highly susceptible growth from the Požegača scion in the same tree.

Some trees of each variety are left uninoculated. These trees serve as checks. They show the characters of growth and fruit production of a healthy tree for comparison with such characters in trees of the same variety that are carrying sarka virus as a result of graft-inoculation. The uninoculated trees also serve as checks on natural spread of sarka in the experimental orchard.

The results of this research will be of great value in choosing plum varieties and selections for future plantings, and in selecting parent trees for breeding high-quality plum resistant to sarka. One of the best methods for reducing disease losses is to grow varieties or selections that are highly satisfactory and at the same time resistant or tolerant to damaging diseases and disorders. It would appear desirable to subject all plum and prune varieties, local or introduced, to sarka-inoculation tests before planting in locations where it is believed that sarka is undergoing important natural spread. More than 50 varieties or selections of plums were being tested in this way at Sarajevo in the summer of 1956.

A catalog or chart describing symptom manifestation of sarka in different plum varieties would be a valuable aid in scouting for the disease, in estimating losses, in marking diseased trees for removal, and in inspecting nurseries. Pobegajlo, Jordovic, and others have given some thought to this matter. A system of rating severity of the different symptoms in leaves, fruit, and shoot growth could be devised in the form of a simple chart or outline which would provide space for checking off the characters and their intensity, as observed in the variety at the place and the time the record was being made.

It should be pointed out that under some circumstances, use of plum varieties highly tolerant to sarka virus could be of questionable value. Plum trees of all varieties may be assumed to be susceptible to infection by sarka virus, but some varieties will show much more striking symptoms of the disease than others. Infected trees of varieties in which symptoms tend to be obscure are apt to be overlooked in orchard inspections. In areas where sarka is reported to be spreading rapidly, as in Bosnia, trees tolerant to the virus and showing few to no symptoms could escape removal in a campaign to control the disease. If susceptible varieties such as Požegaća are the leading commercial sorts in the area, and if for their protection it is deemed necessary to remove the diseased trees in orchards and nurseries, the symptomless trees of the tolerant varieties could harbor the virus and be very troublesome. Such infected trees could be discovered by indexing, after which they could be destroyed - a time-consuming and somewhat costly procedure where a great many trees would have to be indexed. In some areas it might be preferable to discontinue growing such varieties altogether. In other areas, where sarka is abundant and vectors very effective, the costs of continued inspection and removal of diseased trees could be prohibitive; in such cases, to maintain plum culture the district should consider abandonment of the highly susceptible varieties in favor of varieties that are tolerant of the disease. How a case involving some of these questions was handled in North America is discussed in the following paragraphs.

In the Palisade area of western Colorado, U.S.A., there is a large and very important commercial peach industry in which the Elberta and J.H. Hale varieties are grown for transport to central and eastern markets. The area is famous for its Elberta and J.H. Hale peaches and these are by far the leading varieties. Among these orchards, however, there were scattered plantings of early-ripening, white-fleshed varieties for local consumption before the crop of Elberta and J.H. Hale peaches matured.

As mentioned previously, peach mosaic (10) appeared in 7 trees in the Palisade area in 1931. Its rapid spread is described in the section of the report dealing with diseased-tree removal. Over 30,000 new cases of peach mosaic appeared in these orchards in 1935. A control campaign lowered the incidence of new cases to about 4,500 by 1937, but for some years thereafter it was impossible to reduce this figure further. Finally, it was discovered that the virus was spreading to the Elberta and J.H. Hale trees from trees of the early-ripening white peaches, which were virtually symptomless carriers of the virus and were being overlooked in the annual inspections for mosaic. Indexing all of these trees to determine which ones were infected would have been a large undertaking. The growers realized that their industry was dependent on the Elberta and J.H. Hale varieties, both of which are extremely susceptible to peach mosaic. It was decided to remove all trees of the early varieties. This was done, removal of diseased trees was continued and the disease was brought under control.

Preventive and curative treatments

Use of chemical therapeutants for prevention or control of virus diseases of fruit trees is still in the experimental stage. Nevertheless, it is a rapidly expanding field of research and progress is being made. For example, Stoddard (21) showed in 1941 that the virus of X-disease of peach in New England could be inactivated by soaking budwood from diseased trees in aqueous solutions of hydroquinone, p-nitrophenol, or several other chemicals. In later experiments, working with sand cultures of small, potted peach trees in a glasshouse, he demonstrated that the X-disease virus could be inactivated by injections of certain sulfa compounds and other chemicals into the trees or by soil applications of various chemicals. Controls of this kind have not yet been developed for orchard and nursery trees. It is suggested that experimentation along these lines be included in the research on sarka disease.

It is well known that heat treatments may be employed to inactivate certain pathogenic viruses in plant tissues. Usually heat is applied by submerging the material in water maintained at temperatures and for time periods found effective against each particular virus.

Reports from Bulgaria and Yugoslavia indicate that sarka virus was inactivated in infected plum scions at about 66°C. It would be desirable to explore such treatments somewhat further and determine whether longer exposures at lower temperatures, which would be less injurious to the buds and other tissues, might be employed. If successful, this method could be used for making such material safe for distribution and planting. In the case of phony disease, it was found that the virus in dormant peach nursery trees could be inactivated by submerging the trees for 40 minutes in water maintained at 48°C. (22).

VII. RECOMMENDATIONS

Résumé of control recommendations

A strong program should be organized for detection and removal of plum trees that manifest sarka disease. Sarka trees carry an infectious virus, occupy valuable land, and give low return; and there is no practical cure for diseased trees in orchards and nurseries. Widespread dissemination of the disease comes about largely through unwitting use of infected propagating material from diseased trees. Insect vectors are reported to spread the virus from diseased to healthy trees in Bosnia. A progressive program of removal of diseased trees would greatly reduce these sources of infection. Coordination of the various activities required by the Republics in a national program of this kind would be most effective if it were a function of a central agency of the Government.

Plum-tree registry, bud-certification service, and virus-free scion orchards should be provided so that nurserymen, fruit growers, and horticulturists have access to approved, healthy propagating material.

Tests to determine the relative susceptibility of plum varieties to sarka are well under way; these should be extended to include the full range of old and new varieties promising for crop production and breeding.

The possible relation of insect vectors to spread of sarka is of great importance. Tests in which transmission was accomplished by aphids should be repeated on a large number of test plants, and the research should be extended to provide an appraisal of the disease spread by vectors under field conditions in the several Republics. Where practical, vector control should be applied to reduce spread of the disease.

Preventive and curative treatments for fruit-tree virus diseases are in the experimental stage of development, and show some promise. Efforts in this direction should be part of the research program for sarka. Chemotherapy of diseased trees, along with heat treatments for inactivating the virus in dormant nursery trees, suckers, and scions, offer important fields for investigation.

Research institutions

The Institute of Fruit Culture at Cacak and the Faculty of Agriculture at the University of Sarajevo are conducting very important research on sarka and they have begun studies on other virus diseases of fruit trees. The two institutions complement each other in research, and their programs are coordinated in such a way as to make good use of their facilities. Virus diseases of cherries are being investigated at the Institute of Plant Culture, at Zagreb.

The Institute of Plant Pathology of the Faculty of Agriculture, University of Belgrade, is in the process of organizing a special laboratory for research on plant virus diseases and for training students in this very important field.

The Pasteur Institute of Novi Sad and the Faculty of Agriculture, University of Novi Sad, Serbia, are laying plans for a cooperative program of research on viruses and virus diseases in the fields of medicine, veterinary science, plant pathology, and entomology. This is a highly significant program and holds much promise for real advancement in the far-reaching and increasingly important field of virus diseases. Few institutions in the world as yet have undertaken this much-needed, unified approach to virus research.

Facilities and equipment. At the outset, facilities and equipment for virus research at these institutions should be adapted to the program requirements. The whole history of virus research shows that with small outlay for equipment, a trained observer may acquire a great deal of pertinent information about a plant disease known or suspected of being caused by a virus.

When the virus nature of a disease is determined, then there is place for experimental nurseries and field plots. As virus research develops, facilities for isolating plants will be required. Glass-houses are an important adjunct not only for plant propagation but also for doubling the effective working time of the investigator. Finally, laboratories where botanical, plant pathological and biochemical investigations can be pursued will be needed.

Experimental nurseries, orchards, and field plots. Experimental and observational areas provide a proving ground for determining whether disease symptoms in fruit trees are associated with a transmissible virus or are due to some other cause. Abnormalities simulating the symptoms of virus diseases are very common in the foliage and shoot growth of fruit trees. Such abnormalities frequently result from mechanical injuries, nutritional deficiencies and excesses,

unfavorable weather, genetic disturbances, and injuries due to fungi, bacteria, or insects. In survey and control work it is necessary to become familiar with the true and the false symptoms of diseases. When recognition of symptoms is doubtful, graft-transmission tests should be freely employed on healthy trees in the "proving ground".

Additional uses of these experimental and observational areas are many, including: (1) For determining the host range of a virus; (2) for comparing the character and severity of disease symptoms in the leaves, fruits, and shoot growth of different varieties of plum, peach, or other species of fruit trees following inoculation of the trees by grafting; (3) for evaluating the effects of a disease on size and quality of the crop; (4) for computing tree losses and the rate of spread of a disease; (5) for discovering the resistant or tolerant hosts among cultivated varieties and related wild species; (6) for ascertaining the symptomless carriers of a virus; (7) for observing whether there are some strains of a virus that produce mild symptoms and others that produce severe symptoms, and whether the mild strains will immunize a tree against the severe strains; (8) for determining the incubation period of a disease; (9) for vector research; (10) for demonstrating whether a virus is carried in the seeds; (11) for showing the response of diseased trees to chemical fertilizers and pruning; and (12) for chemotherapy and other preventive or remedial treatments.

Knowledge of host range and varietal susceptibility is of immediate importance in connection with any virus disease. A great deal of time can be saved and control procedures can be more advisedly administered if this information is obtained promptly. Healthy trees representative of the most important varieties should be grown in abundant supply on the "proving ground" and should be ready for use in inoculation tests when needed. Just as the bacteriologist and the mycologist characterize and describe organisms by their behavior on a selected range of culture media, so must the virologist employ living trees as the test tubes for his study of fruit-tree viruses and the diseases they cause.

Among the living-tree "test tubes" in Yugoslavia should be included species and varieties that have been used for proving virus diseases of fruit trees in other countries. By inoculating these differential hosts it can be determined whether a newly discovered disease is identical with or similar to some virus disease already described abroad.

There are many virus diseases of stone fruits which have been described from other parts of the world but are not known to occur in Yugoslavia, some of them being devastating and ruinous. Those diseases are apt to find their way to Yugoslavia. By all means the Yugoslav pathologists should be prepared to cope with them when they arrive. They should know what to look for, know how to recognize the diseases, and be equipped to perform confirmatory inoculation tests when suspicion points to an unwelcome introduction of virus origin.

If the tests are positive, immediate action is advisable. No time should be lost in waiting to see whether the disease will undergo natural spread near its point of introduction in the country. It is safer to assume that a natural vector of the virus may be present or soon may be introduced.

An effort should be made to ascertain where and how the disease gained entrance into the country. The area should be surveyed thoroughly and inspections made farther afield. It frequently happens that distribution is much more extensive than at first suspected. Every identifiable case of the disease should be destroyed except those closely isolated and needed for research. If promptly and thoroughly carried out, this program may be expected to prevent or retard further important spread of the new disease, and to hold losses to a minimum.

The experimental nurseries, orchards, or field plots should be located near where the disease is known to occur. This is done to guard against the hazard of accidental introduction of a virus into new areas through the infected propagating material employed in the transmission tests.

Hosts for identifying stone-fruit virus diseases. The importance of having suitable host trees available for identification of fruit-tree virus diseases was mentioned in the preceding section. In some cases it may be a question of confirming identification of a disease known to occur elsewhere in the country. In other cases it may be a question of determining whether a disease recently found in Yugoslavia is the same as a disease described in a foreign country or an entirely new one. An imposing number of virus diseases of Prunus species are known in America and an increasing number is being reported elsewhere.

Most of the stone-fruit virus diseases that occur in North America were described and illustrated in Agriculture Handbook 10 (20). Diagnostic hosts of these diseases have been selected by a Host Range Committee of the U.S. Inter-regional Stone-Fruit Virus Group. This

Committee, under chairmanship of L. C. Cochran, prepared a report entitled "Hosts for Identifying Stone Fruit Virus Diseases", dated 1 December 1950. The hosts recommended by the Committee and the diseases these hosts will identify are shown in table 2 adapted from this report.

Table 2. Diagnostic hosts for stone fruit virus diseases

Name of host		Diseases manifested
Species	Variety	
1. Peach (<u>Prunus persica</u>)	Elberta.....	Peach yellows
		Little peach
		Red suture
		Peach rosette
		Rosette mosaic
		Phony
		Peach mosaic
		X-disease
		Western X-disease
		Yellow bud mosaic
		Wart
		Peach mottle
		Peach necrotic leaf spot
		Asteroid spot
	Golden net	
	J. H. Hale.....	Peach calico
		Peach blotch
Ring spot		
Muir.....	Willow twig	
	Muir peach dwarf	
2. Sour cherry (<u>P. cerasus</u>)	Montmorency.....	Sour cherry yellows
		Green ring mottle
		Necrotic ring spot
		Pink fruit
		Peach mottle
2a. Sour cherry	On mahaleb rootstock	Western X-wilt and decline

Table 2. Diagnostic hosts for stone fruit virus diseases
(Continued)

Name of host		Diseases manifested
Species	Variety	
3. Cherry, sweet (<u>P. avium</u>)	Bing.....	Buckskin Albino Mottle leaf Rusty mottle Mild rusty mottle Rasp leaf Twisted leaf Tatter leaf Small bitter cherry
	Royal Ann..... (Napoleon)	Peach mottle Black canker Sweet cherry rugose mosaic Pinto leaf
	Lambert.....	Necrotic rusty mottle Little cherry Small bitter cherry Lambert mottle Utah Dixie rusty mottle
	Shiro-fugen.....	Ring spot
	Kwanzan.....	Other latents Rough bark
	Italian Prune.....	Prune dwarf
	French (prune).....	Prune diamond canker
	Standard (prune)	Standard Prune constricting mosaic
	Shiro.....	Line pattern
	Santa Rosa.....	Plum white spot
4. Cherry, oriental flowering (<u>P. serrulata</u>)	Abundance and Burbank	(a)
5. Plum, European (<u>P. domestica</u>)		
Plum, Japanese (<u>P. salicina</u>)		
6. Apricot (<u>P. armeniaca</u>)	Tilton.....	Ring pox
7. Almond (<u>P. amygdalus</u>)	Nonpareil.....	Drake almond bud failure

(a) These two plums appear to be useful as screening hosts for eliminating ring spot and sour cherry yellows from other virus cultures to which they are hosts.

The Host Range Committee later was given the tasks (1) of selecting a minimum list of test plants for indexing foreign importations of stone fruits into the United States, for presence of viruses, and (2) of locating sources of virus-free materials for propagating the test plants to be used. Indicator plants proposed by the Committee in a report dated 30 November 1954, together with the Committee's later recommendations concerning scion-rootstock combinations for virus-free test plants, are shown in table 3. It is recognized that this list is not sufficient to indicate some of the known stone-fruit viruses: for example, Muir peach dwarf, prune diamond canker, and Lambert mottle. However, so far as present knowledge goes the list is sufficient for diagnosing the more important stone-fruit virus diseases that occur in North America and elsewhere.

Table 3. Scion-rootstock combinations
for virus-free test plants

Indicator scion	Rootstock
Elberta peach	Lovell peach seedling
Nonpareil almond	Lovell peach seedling
Tilton apricot	Lovell peach seedling
Italian Prune	Lovell peach seedling
Shiro plum	Lovell peach seedling
Montmorency cherry	Mahaleb cherry (<u>P. mahaleb</u>)
Bing cherry	Mahaleb cherry
Shiro-fugen oriental flowering cherry	Mazzard cherry (<u>P. avium</u>)

Indexing of stone fruit importations into the United States is done by the New Crops Research Branch, Agricultural Research Administration, United States Department of Agriculture. Sources of test-plant or indicator-plant material are selected with great care. Upon request to Howard L. Hyland, in charge of the Plant Introduction Section of the New Crops Research Branch, a limited supply of virus-free scions from trees of the species and varieties employed as test plants could be sent to Yugoslavia. With this material as a foundation, the respective test plants could be multiplied locally by propagation on stocks known to be virus-free.

Glasshouses and screened enclosures. Greenhouse is commonly used for plant propagation and is especially adapted for rooting fruit-tree cuttings, which is best done in sand over bottom heat such as can be provided in a glasshouse. Modern methods permit the breaking of dormancy, so that plants can be started in the glasshouse and investigations can proceed as effectively in winter as in summer, thus doubling the effective working time of the pathologist.

For detailed virus studies, the facilities of a glasshouse are especially valuable. Separate compartments should be provided (a) for growing healthy plants to be used in inoculation experiments; (b) for maintaining cultures of plants infected with a virus under study, which serve both for demonstration of disease symptoms and for supplying inoculum; (c) for maintaining pure cultures of insects to be tested as possible vectors; and (d) for performing tests employing insects that have fed on diseased plants or on juice or tissues from diseased plants.

A glasshouse for these purposes should be well screened and should be equipped for regulating heat, humidity, light, and ventilation, as may be required for good development of test plants and insect vectors. Smaller, portable, glass-walled chambers with automatic control of temperature and humidity may be placed within large compartments, when precise regulation of environmental conditions is desired.

Before and after inoculation some of the experimental plants may be grown satisfactorily in screened enclosures out-of-doors. A natural environment is provided in this way, and at the same time the plants are protected against unwanted insects and other pests. Crowded conditions in the glasshouse may be relieved, where practicable, by moving potted test plants from the glasshouse to screened, outdoor enclosures.

Smaller houses with aluminum frames are now available in America at low cost, permitting the setting up of a series of small houses at the same cost as one house of standard construction. Important developments also have taken place in the use of plastics to replace glass.

Laboratories. Facilities such as are commonly available in laboratories for research on plant diseases and insect vectors should be maintained at the fruit-tree virus research centers. These should include the usual equipment such as compound and dissecting microscopes, microtomes, glassware, reagents and equipment used in microtechnique, juice-extraction apparatus, chemical and biochemical equipment and reagents, electric ovens, incubator, refrigerator, and centrifuge.

Equipment should be on hand for rapid drying of specimens showing mottles and deformities in leaves and twigs, for mounting the dried specimens, and for storing the labeled specimens in herbarium cases. It is particularly important to have available good equipment for making detailed photographs, black and white and colored. Transmitted light will be needed to bring out in photographs some of the color patterns in leaves from diseased trees. Cases and files for negatives, prints, and color transparencies will be needed. A table viewer and a standard projector and screen for lantern slides and color transparencies should be provided.

The newer developments in biochemistry seem to have considerable value in virus research. This is particularly true with chromatography, which permits by the use of relatively simple apparatus the determination of the presence of various plant constituents such as carbohydrates, amino acids, alkaloids and other substances, and at the same time permits their quantitative appraisal.

Access to an electron microscope will be desirable for some phases of virus research. This will be particularly the case at the new virus laboratory to be constructed by the Faculty of Agriculture at the University of Belgrade, where the broad field of plant virology will be studied and taught. Expensive equipment of this kind would have much less use in studies of viruses that attack fruit trees, since very few of these viruses have been shown to be juice transmissible by ordinary techniques.

Service headhouse. Provision should be made for storing, mixing, and sterilizing soils, for cleaning and sterilizing soil containers, for potting plants, and for burning discarded plant material. A service headhouse is useful for these purposes. A convenient arrangement is to have the laboratory, the glasshouse, and the service headhouse connected by doors or corridors, which make the units accessible to the workers and facilitate moving plants from one place to another.

Conferences and meetings

Conferences within the country. Investigations and control of fruit-tree virus diseases in Yugoslavia would be advanced by holding annual conferences among workers and officials engaged in these activities. Pathologists, entomologists, horticulturists, station directors, and representatives of the Federal Government and the Republics could profitably meet for discussion of the problems, of progress made with program activities, and of future needs in research, survey, and control. Some of these meetings could be in the nature of field trips in summer so that those attending could become acquainted first-hand with recent developments and see the diseases under field conditions.

International meetings. Also, Yugoslavia should be well represented at important meetings held elsewhere in Europe for discussion of research and control of fruit-tree virus diseases. It is recommended that one or more pathologists engaged in research on sarka and other virus diseases of fruit trees in Yugoslavia participate in the Fourth Symposium on Fruit Tree Virus Diseases in Europe, which will be held at Copenhagen, in 1960.

Study and travel in foreign countries

The Government of Yugoslavia has made a commendable effort to secure training privileges for some of its promising young pathologists at leading institutions outside the country. Such travel is particularly important for fruit-tree virologists. Publications on virus diseases are important, but they cannot take the place of well-timed visits to confer with virologists at foreign research institutes, to see the diseases first-hand, and to become familiar with procedures having to do with research, surveys and control undertakings.

It is recommended that travel for these objectives should include some countries of the Western Hemisphere. Particularly, it would be advantageous to visit the United States and Canada, which countries were the first to investigate a fruit-tree virus disease and to apply effective control measures.

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